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Physiological and sanitary potential of peanut seed treated with cinnamon powder

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This study aimed to assess the physiological and sanitary potential of peanut seeds cv. BR1, treated with powdered cinnamon (*Cinnamomum zeylanicum*). The experiment was conducted in the laboratories of the Seed and Plant Pathology and Microbiology at the Federal Rural University of Pernambuco / Academic Unit of Garanhuns (UFRPE / UAG). The experimental design was completely randomized, with four replications of 50 seeds. The treatments consisted of treating the seeds with cinnamon powder at different doses (0: control; 2, 4, 6, 8 and 10 g) in the ratio of 2 g powder and 81.2 g. The seeds were soaked in 1% sodium hypochlorite for 3 min, and then immediately subjected to the following tests: germination, speed of germination index, seedling length, seedling dry matter and seed health testing. Fungi of the genus *Aspergillus*, *Penicillium* and *Rhizopus* were detected greatly in the treatments with the highest dose. Sodium hypochlorite was effective in combating fungal growth, but damaged the physiology of the seeds. Using cinnamon powder of 5 g was efficient in controlling fungi and did not affect the seed quality of peanuts.

Key words: *Arachis hypogaea*, natural fungal control, *Cinnamomum zeylanicum*.

INTRODUCTION

Peanut (*Arachis hypogaea* L.) belongs to the family Fabaceae; subfamily Papilionidae, and gender *Arachis*. The economic importance of peanuts is linked to their having pleasant taste and seeds having rich oil (40 to 50%) and protein (22 to 30%) (Silveira et al., 2011). In Brazil, production is more significant in São Paulo, which is the largest producer; it accounted for 96.67% of the country's total production in the 2012/2013 crop season, followed by Bahia State, with 3.33% (Conab, 2013). In

the semiarid region of Pernambuco State, it has been tested that doing intercropping with cactus leads to achieving high yields (Andrade et al., 2015).

Peanut crop is propagated by seeds, which constitutes a major component production. It is of great importance to maintain the quality of seeds used in the implementation of this culture, especially regarding its genetic, physical, physiological and phytosanitary attributes (Grigoletto et al., 2012). Seed quality is of

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utmost importance in the production process of any plant species, because it influences the development of culture.

The physiological quality of peanut seeds can be compromised, and even lost gradually. This can happen through deterioration influenced by fungus contamination (especially those of the genus *Aspergillus*, *Penicillium*, *Fusarium* and *Rhizopus*) during the stages of maturation and harvest (Pitt et al., 1991), post-harvest, drying (Fernandez et al., 1997) and processing (Almeida et al., 1998).

In conventional production, seeds are often treated with chemical fungicides. This reduces seed and seedling losses caused by seed-borne and soil-borne diseases. Most seed protectants are not an option for organic growers. Herbal and plant extracts treatment and the use of hot water can be used by organic farmers to improve seeds' performance. It is therefore necessary to search for control measures that are cheap, ecologically sound and environmentally safe to eliminate or reduce the incidence of these economic important pathogens, so as to increase seed germination.

The losses caused by pathogens has been reduced, mainly by the use of chemicals, either pre- or post-harvest, however, the use of agrochemicals has led the emergence of numerous high severity problems such as the emergence of microorganisms resistant to fungicides, environmental damage and harm to human health (Cruz et al., 2009). Alternatively, the use of pesticides has been researched as natural products. Shah et al. (1992) reported that *Argemone mexicana* seed extract was effective in eliminating most of the seed-borne fungi of cowpea but was not effective against *Alternaria alternata*, *Curvularia lunata*, *Mucor* sp. and *Macrophomina phaseolina*.

Some plants have in their chemical composition active substances that can act as fungitoxic in plant-pathogen interactions with other antimicrobial activities; they can even activate the defense mechanisms of the host plant with pathogen control programs, thus reducing the indiscriminate use of pesticides (Silva et al., 2011).

In this sense, medicinal plants have been extensively studied for the control of various pathogens, due to the active principles present with potential fungicidal and / or bactericidal. Among them, we can mention Cinnamon (*Cinnamomum zeylanicum* L.); in addition to its use as a spice or condiment for providing flavor and aroma in food, it has fungitoxic properties; the main antimicrobial constituent, cinnamaldehyde (Ranasinghe et al., 2002).

There is a growing need to evaluate the sanitary quality of peanut seeds, in order to select highly physiological quality and pathogen free ones. Thus, this study aims to evaluate the physiological and sanitary quality of peanut seeds cv. BR1 treated with cinnamon powder.

MATERIALS AND METHODS

The experiment was conducted at the Seed Analysis Laboratories

and Plant Pathology and Microbiology, Federal Rural University of Pernambuco / Academic Unit of Garanhuns (UFRPE/UAG), Pernambuco State. The peanut seeds cv. BR1 used were purchased from Embrapa Cotton - Campina Grande, Brazil.

At the Seed Analysis Laboratory, the peanut cultivar BR1 seeds were treated as follows: the control (0) and the experimental seeds were immersed in sodium hypochlorite for 3 min, according to Brasil (2009), using different doses of cinnamon powder (*Cinnamomum zeylanicum* L.): 2, 4, 6, 8 and 10 g. Seeds treated with cinnamon powder dose remained conditioned for 24 h in a plastic bag together with dust. Treatment with 1% sodium hypochlorite solution was done according to the recommendations given for Sanitary Analysis Manual seeds (Brasil, 2009). To obtain the hypochlorite 297 mL of distilled water was mixed with 3 mL of sodium hypochlorite. After the treatments, there was the germination and health test.

Physiological quality test

The germination test was performed according to guidelines of the Rules for Seed Analysis-RAS (Brasil, 2009), using germination chambers type Biochemical Oxygen Demand (B.O.D), equipped with fluorescent lamps. It was done under constant temperature of 30°C in substrate, Germitest brand; it was sterilized for 20 min at 105°C ± 3°C and moistened with distilled water. The amount was equivalent to 2.5 times the weight of the paper.

The percentage of germination was carried out at 10 days after sowing. Seedlings with well-developed essential structures were considered as normal (Brasil, 2009).

First count

First count was held on the fifth day after the beginning of the experiment, corresponding normal seedlings.

Speed of germination index

Speed of germination index was carried out simultaneously with the germination test; daily counts were done from the fifth day. At the end of the test, the index was calculated using the formula:

$IVG = \frac{G1}{N1} + \frac{G2}{N2} + \dots + \frac{Gn}{Nn}$, where IVG = germination rate index; G1, G2 and Gn = number of normal seedlings, computed in the first, second... and last count, respectively; N1, N2, Nn = number of days from sowing to first, second... and last count, respectively (Maguire, 1962).

Seedling length

Seedling length seedlings were measured on the 10th day after sowing, using a ruler graded in centimeters, measuring from the root to the epicotyl; the result was divided by the number of seedlings.

Dry mass of seedlings

Seedlings were wrapped in paper bags without the cotyledons, put in oven at 80°C for 24 h, and then weighed on an analytical balance; the result is expressed in grams (Nakagawa, 1999).

Health test

The seeds were distributed on two sheets of paper germitest,

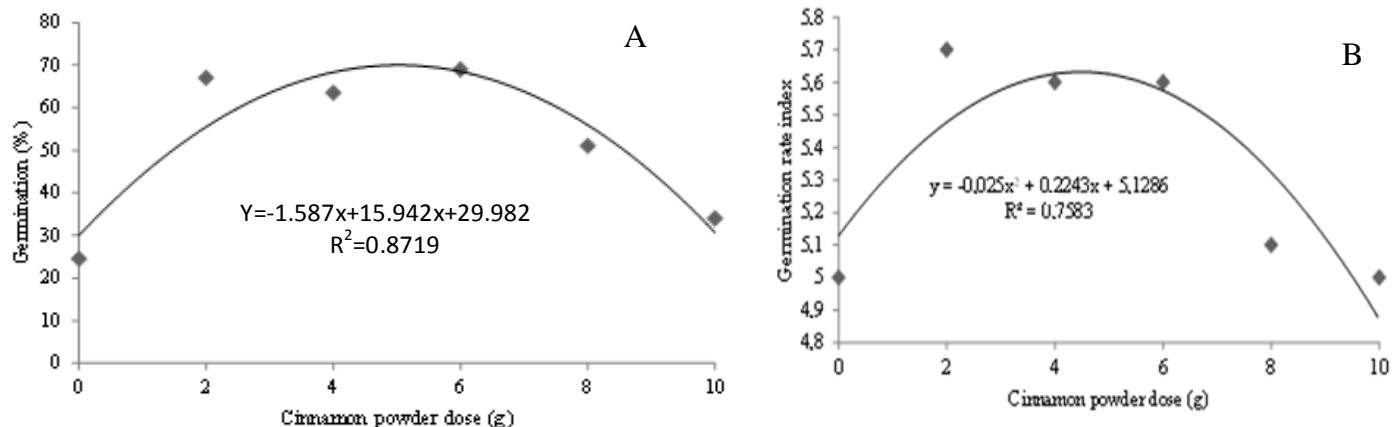


Figure 1. Germination (A) and germination speed index (B) of *Arachis hypogaea* seeds treated with cinnamon powder (*Cinnamomum zeylanicum* L.).

sterilized for 20 min in an oven at $105 \pm 3^\circ\text{C}$ and moistened with distilled water (control) and the concentrations of the extract. The amount is equivalent to 2.5 times the weight of the paper. The rolls were maintained in a growth chamber (B.O.D.), equipped with fluorescent lights at 20°C temperature. The test duration was 8 days; the evaluations were done on the eighth day after the beginning of the experiment, according to the rules of Sanitary Analysis Manual Seeds (Brasil, 2009).

To identify and quantify the percentage of fungi, each seed was visualized with the aid of a magnifying glass (60 times magnification) and an optical microscope, allowing the detection of fungal structures. The aggregation of fungi was performed by percentage of seeds contaminated, resulting in the average per treatment.

For physiological and health quality tests, we used the completely randomized design with four replications of 50 seeds, totalling 200 seeds per treatment. The data of the variables measured in the experiment were subjected to analysis of variance using the F test and the means compared by Dunnett "t" test at 5% probability. Quantitative data was submitted to polynomial regression analysis, testing the linear and quadratic model, choosing the highest R^2 . The analyses were done using the software SAEG (Ribeiro, 2001).

RESULTS AND DISCUSSION

Figure 1 shows the results of germination (A) and germination rate index (B) on *Arachis hypogaea* seed cv. BR1 treated with doses of cinnamon powder. When the seeds were treated with a dose of 5.0 g of powder, there was an increase in the percentage of germination (71%); the highest rate was 5.6 at a dose of 4.5 g. It can be seen that the treatment of seeds with cinnamon powder did not impair germination and germination speed index. Ferreira and Áquila (2000) found that germination is less sensitive to allelochemicals compared to the growth of seedlings. The better performance of treated seeds compared to the control is because cinnamon powder is reliable to control fungi.

The answer to the interference of allelochemicals on

seed germination depends on the concentration used (Ciarka et al., 2002). Furthermore, the influence of the type of extract also depends on the sensitivity of the tested plant to allelopathic compounds present; with certain substances it can inhibit germination or growth, and with others it can be the same or stimulating innocuous (Almeida et al., 1998).

This study confirms that natural mycoflora present in seeds of *A. hypogaea* was capable of causing poor seed germination and had negative influence on seedling growth. *C. zeylanicum* L. increase germination and germination speed index in some doses. This could be attributed to the suppression of the incidence of the seed borne fungi that could have killed the embryo of the seeds. This result is consistent with that of Parimelazhagan and Francis (1999) who established that leaf extracts of *Clerodendrum viscosum* increased seed germination and improved seedling development of rice seeds.

Cinnamon has a chemical composition in the presence of substances such as cinnamic aldehyde (75-90%), aldehyde benzoic acid, cinnamic acid, coumarin, methyl-ortocumaraldehyde, salicylic acid methyl ester to cinnamic acid (Gerhardt, 1973). The cinnamaldehyde is the main component in fungal activity present in cinnamon extracts. Other chemicals have additive or synergistic effect of totally fungitoxicity (Jham et al., 2005).

The values for the length (A) and seedling dry weight (B) derived from peanut seeds treated with cinnamon powder are shown in Figure 2. It was observed that when the seeds were treated with 5.2 g of powder, there was the greatest seedling length of 7.7 cm and the greatest value for the dry mass was 0.025 g at a dose of 4.8 g. That dose reduced early growth and dry mass. Probably, this interference is related to the high concentration of allelochemicals present in cinnamon powder, as cinnamic acid. The compound (E) - cinnamaldehyde (97.7%) was

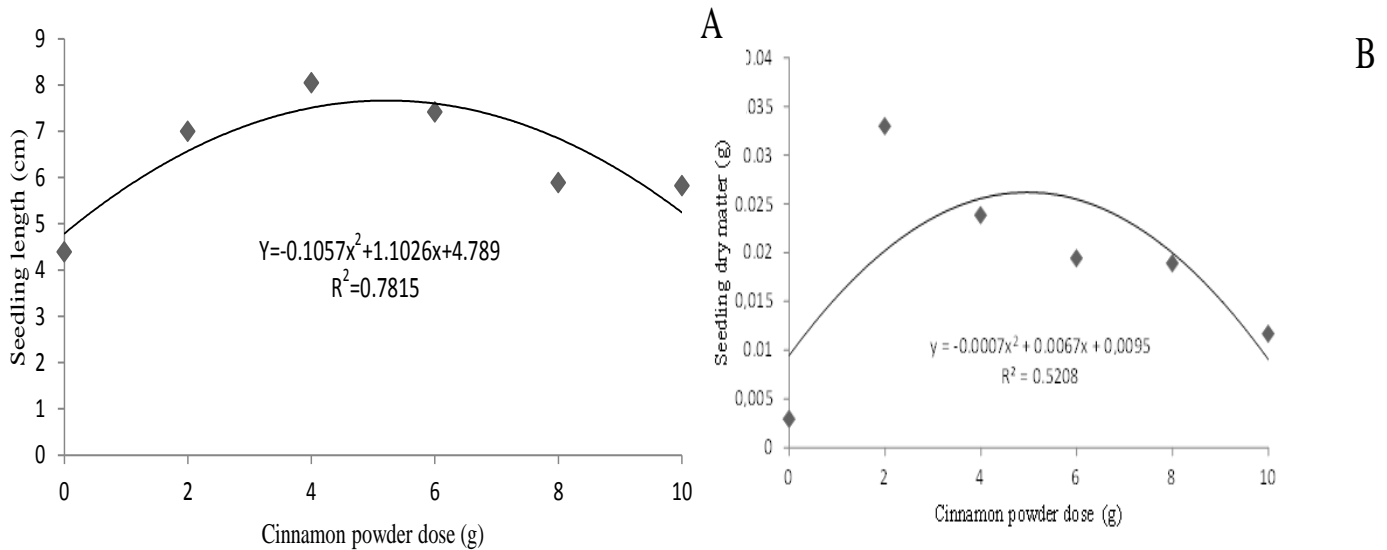


Figure 2. Length (A) and seedling dry weight (B) of *Arachis hypogaea* treated with powdered cinnamon (*Cinnamomum zeylanicum* L.).

found in large amounts (97.7%) in the essential oil of *C. zeylanicum*, being considered as the main component (Singh et al., 2007). The initial growth phase appears to be more sensitive to allelochemicals than germination, which may lead to abnormal seedlings, root necrosis, or alterations in the growth (Ferreira and Borghetti, 2004). The influence of natural extracts on the growth of seedlings shows the ability of compounds acting at this stage of development (Lousada et al., 2012). The potential of natural products to inhibit the growth of seedlings may be related to the chemical constituents present in the plant or the interaction of its components (An et al., 1993).

The results obtained by Nwachukwu and Umechuruba (2001) showed that crude extracts from all the plant leaves tested increased seed germination, led to the emergence of African yam bean seeds and gave significant ($P \leq 0.05$) reduction of mycelial growth of all the fungi tested compared to their aqueous extracts. Leaf extracts of neem, basil, bitter leaf and paw-paw, which are cheap and environmentally safe, are promising for protecting African yam bean seeds against major seed-borne fungi and can also improve the crop. Perelló et al. (2013) suggest that seeds treated with garlic juice had a relatively better germination percentage, plumule and radicle length, and seedlings than those seeds treated with high inoculum density of the target pathogen, *Bipolaris sorokiniana*. The inoculum level of naturally infected wheat seeds could be reduced through the use of garlic juice as seed dressing biofungicide, before sowing. Kiran et al. (2010) and Yassin et al. (2012) recorded the efficacy of the antifungal properties of some herbaceous and medicinal plants against cereal seed-borne mycoflora. The variation in results related to seedling growth can be explained by the fact that there is

a pattern of response to the concentration. Certain extracts even at low concentrations can interfere with growth of some species and not others. Hence, there is need to always perform specific tests involving species of plants and extracts concentrations (Premasthira and Zungsontiporn, 1996).

Figure 3 shows the results for the control of phytopathogens using peanut seeds treated with cinnamon powder. The use of cinnamon powder proved to be efficient in controlling these types of fungi (*Aspergillus niger*, *Penicillium* spp. and *Rhizopus* spp.); it led to a marked decrease in the percentage of these fungi with increasing doses. This result agrees with the findings of Zaman et al. (1997), that the efficacy of garlic, neem, ginger and onion extracts on seed borne fungi of mustard declined with increased dilution.

Similar results were found by Mamprim et al. (2013) on plant extracts of cinnamon, rue (*Ruta graveolens*), cinnamon (*Melia azedarach*), rosemary (*Rosmarinus officinalis*), lemon grass (*Cymbopogon citratus*) and citronella (*Cymbopogon winterianus*), which caused a reduction in diameter growth of *Metarhizium anisopliae* (Metsch.) Sorok from 1.8 to 7.6%. The essential oils of cinnamon and oregano (*Origanum vulgare*) at a concentration of 100 ppm have antifungal effect against *Aspergillus flavus* (Camarilho et al., 2006).

The results observed in the physiological quality test (Figures 1, 2 and 3) indicate that the control treatment showed lower seed behavior for germination and vigor compared to those treated with cinnamon powder. This can be justified by the presence of fungi in seeds untreated. The incidence of seed-borne pathogens can affect the physiological quality and, in some cases, inhibit seed germination. It is therefore necessary to search for control measures that are cheap, ecologically sound and

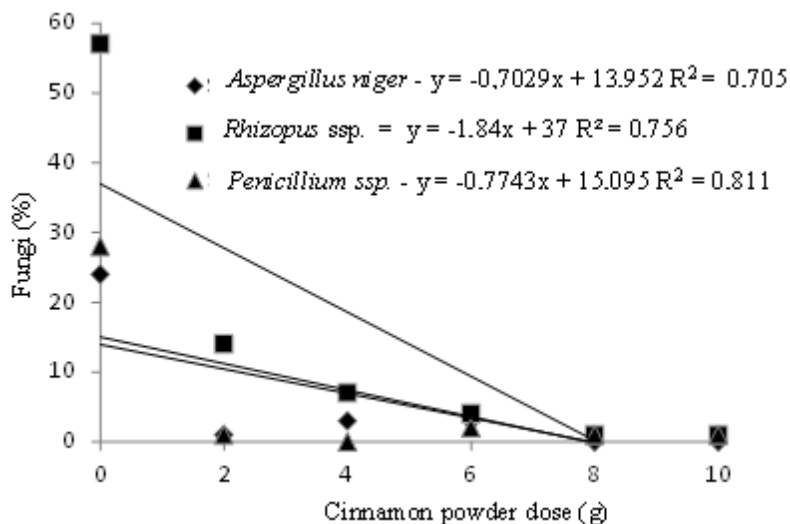


Figure 3. Percentage of fungi in *Arachis hypogaea* seeds treated with doses of powdered cinnamon (*Cinnamomum zeylanicum* L.).

environmentally safe to eliminate or reduce the incidence of these economic important pathogens, so as to increase seed germination.

Chavan and Kakde (2008) reported that fungi such as *A. niger*, *Aspergillus flavus*, *Alternaria dianthicola*, *Curvularia lunata*, *Curvularia pellescens*, *Fusarium oxysporum*, *Fusarium equiseti*, *Macrophomina phaseolina*, *Rhizopus stolonifer*, *Penicillium digitatum* and *Penicillium chrysogenum* cause discoloration, rotting, shrinking, seed necrosis, loss in germination capacity and toxification to oil seeds. *Aspergillus* is a common mould in tropical and sub-tropical countries and causes aflatoxin contamination as a result of moulding of badly stored commodities, such as groundnut, cereal and cotton seeds.

Saha et al. (2014) concluded that seeds like garlic tablet, allamanda tablet or neem leaf extract can be recommended for okra seed treatment to get higher germination and healthy seedling that will eventually produce more fruit.

Aspergillus spp. and *Penicillium* spp. are storage fungi. The interaction of seeds with these fungi can significantly increase the speed of reducing the quality of seeds (Neergaard, 1979). Also, the genus *Rhizopus* spp. is also considered a storage fungus that hinders the identification of other pathogens present in peanut seeds, due to its rapid growth. It covers the seed surface. The growth of storage fungi in seeds can cause death of the embryo, reduce twinning, overturning and cause rot (Mazzani and Layrisse, 1992; Bhattacharya and Raha, 2002). Thus, assessment of health quality is extremely important to check the quality of seeds.

Several studies have been developed in the search for natural products with fungitoxicity and their application in the control of fungi that cause damage to crops of

economic interest (Hillen et al., 2012). Among these products is cinnamon. Cinnamic aldehyde is present in high concentration (77.72%) of cinnamon; it is the main chemical component, and is primarily responsible for inhibiting the growth of pathogenic micro-organisms (Andrade et al., 2012).

Table 1 shows the results of the variables analyzed for germination, as well as the vigor and health of peanut seeds immersed in sodium hypochlorite and those treated with cinnamon powder doses. The first count, the germination speed index and the germination of treated peanut seeds showed significant effects compared to those seeds immersed in sodium hypochlorite solution 1%, for 3 min; except for those treated with 10 g, they were no significant differences. In addition, we observed higher number of abnormal seedlings when seeds were immersed in sodium hypochlorite and higher number of normal seedlings from seeds subjected to doses of cinnamon powder.

These results indicate that cinnamon powder doses were superior to the control and sodium hypochlorite treatment; and even though the use of sodium hypochlorite is a method recommended by RAS, it impaired the germination of peanut seed; this is presumably because soaking causes damage to seed membranes. Evangelista et al. (2007) state that the membranes can suffer from injuries when they are immersed in water when the seeds have water content less than 12%. The first soaking is considered critical because there is a rapid release of electrolytes to achieve a balance in the membranes. In that work, the peanut seeds were under 10% humidity (Kraft, 1997). The seeds of this species have a high degree of susceptibility to injury caused by rapid soaking through immersion in distilled water.

Table 1. *Arachis hipogaea* seeds treated with cinnamon powder dose (*Cinnamomum zeylanicum*) and immersed in sodium hypochlorite (1%).

Treatment	Control (sodium hypochlorite)									
	FC (%)	G%	GSI	LS	DS	ABS	SN	AS	PEN	RIZ
2 (Control)	22.0*	-9.00 ^{ns}	1.39*	1.29 ^{ns}	-0.001	18.0*	-9.00*	43.5*	26.5*	14.5*
3 (Dose 2 g - NaHCl)	27.5*	33.5*	2.03*	1.30 ^{ns}	0.028*	-18.0*	-15.0*	4.00 ^{ns}	-1.50 ^{ns}	-1.5 ^{ns}
4 (Dose 2 g - NaHCl)	25.5*	30.0*	1.81*	2.35*	0.019*	-16.5*	-13.5*	4.00 ^{ns}	-2.00 ^{ns}	-0.5 ^{ns}
5 (Dose 4 g - NaHCl)	25.5*	35.5*	1.88*	1.72*	0.015*	-21.0*	-14.5*	-2.00 ^{ns}	-0.00 ^{ns}	-0.0 ^{ns}
6 (Dose 6 g - NaHCl)	16.5*	17.5*	1.15*	0.19 ^{ns}	0.014*	-10.5 ^{ns}	-7.50*	0.00 ^{ns}	-1.50 ^{ns}	-1.5 ^{ns}
7 (Dose 8 g - NaHCl)	21.5*	0.50 ^{ns}	1.06*	0.13 ^{ns}	0.007 ^{ns}	4.50 ^{ns}	-5.00 ^{ns}	-1.00 ^{ns}	-1.00 ^{ns}	-2.0 ^{ns}

FC, First germination count; G, germination; GSI, germination speed index; SL, seedling length; DS, dry seedling mass; SN, normal seedlings; ABS, abnormal seedlings; AS, percentage of *Aspergillus niger*; PEN, percentage of *Penicillium* spp.; RIZ, percentage of *Rhizopus* spp. ns and * = not significant, * significant at the 5% probability by Dunnett's test.

For coffee beans (*Coffea arabica* L.), Sofiatti et al. (2009) found that seed soaked in sodium hypochlorite at concentrations of 4, 5, and 6% caused a reduction in seed germination and vigor.

For the length of peanut seedlings, only 8 and 10 g cinnamon powder was superior to sodium hypochlorite treatment. The dry weight of seedlings from seeds treated with cinnamon powder dose showed significant results, except for the dose of 10 g; and when compared to seedling dry matter from the control with those of hypochlorite sodium, it was observed that, despite having a significant effect, bleach gave the best result. This is due to pathogen control.

Conflict of interests

The authors did not declare any conflict of interest.

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